AQUACULTURE AND MARINE BIOTECHNOLOGY IN CHINA

6-19 October 1995

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Research and Scientific Exchange Division
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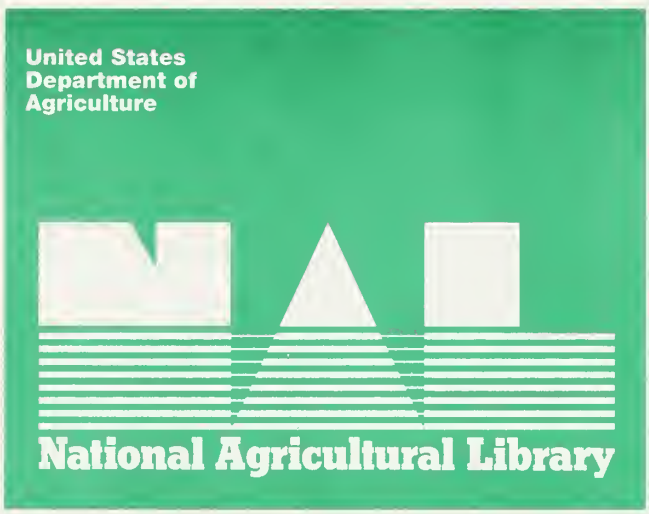
United States
Department of
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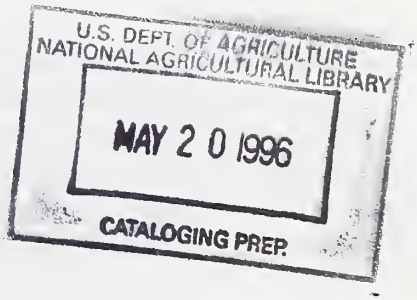
ACKNOWLEDGEMENTS

During our visit in the People's Republic of China, we were hosted by the Ministry of Agriculture. We very much appreciate their efforts that made our visit both pleasant and productive. They were very helpful in establishing our itinerary, making travel arrangements, and providing us comfortable accommodations. The preparations at each location were evident in the excellent presentations, useful discussions, informative tours of laboratory facilities and aquaculture ponds, and exquisite meals. We are particularly appreciative of Wang Donghui, Project Officer of the Ministry of Agriculture's Department of International Cooperations, who traveled with us throughout China. He provided excellent translation where needed. His assistance, as well as that of guides at each city, in providing general information and in helping us cope with a demanding schedule in an unfamiliar culture was critical to the success of this visit.

We also gratefully acknowledge the support of the U.S. Department of Agriculture(USDA), the National Sea Grant College Program of the National Oceanic and Atmospheric Administration (NOAA), the University of Minnesota, and Virginia Polytechnic Institute and State University. The team would also like to acknowledge the support of USDA's International Cooperation and Development, and in particular, Ms. Roslyn Gillespie.



U.S. TEAM VISIT TO CHINA ON BIOTECHNOLOGY APPLICATIONS TO AQUACULTURE

Introduction

A Team of United States (U.S.) biotechnology experts visited the People's Republic of China (P.R.C.), October 6-19, 1995, as part of the U.S.-P.R.C. Scientific Exchange Program administered by the International Cooperation and Development Division, Foreign Agricultural Service, United States Department of Agriculture (USDA/FAS), and the Department of International Cooperation of the P.R.C. Ministry of Agriculture. The purpose of the trip was to survey Chinese aquaculture research and the applications of molecular biology in that research, and to make an overall scientific, economic and ecological assessment of Chinese aquaculture.

The Team was composed of four individuals with diverse disciplinary expertise: Alvin L. Young, Director, USDA/Office of Agricultural Biotechnology (Team Leader); Anne Kapuscinski, Professor, Department of Fisheries and Wildlife, University of Minnesota; Eric Hallerman, Associate Professor, Department of Fisheries and Wildlife Sciences, Virginia Polytechnic Institute and State University; and David Attaway, Program Director for Biotechnology, National Sea Grant College Program, National Oceanic & Atmospheric Administration. A fifth member of the team, William Wolters, Research Leader, Agricultural Research Service (ARS), Stoneville, MS, was forced to cancel due to funding restrictions by ARS. The Team visited location in five cities (Beijing, Shanghai, Wuhan, Shashi, and Guangzhou), which were selected because of their prominence in the published scientific literature on aquaculture research, or on the advice of Chinese colleagues familiar with the research institutions in China. Appendix A provides addresses, goals and objectives for the visit and provides a detailed itinerary and major contacts.

Overview of Chinese Aquaculture and Fisheries

There are more than 500 species of fishes in China. Efforts to collect, identify and provide "type specimens" have occurred at many of the institutions that the Team visited. From examination of the specimens reviewed by the Team, a significant proportion of the specimens were collected in the 1960s through early 1980s. At last fifteen species of fish are valuable in the wild state and 20 are commonly raised in fish ponds. The most popular aquaculture species are the carps and breams (family Cyprinidae) because they grow rapidly to large sizes and accept feed more readily than other species.

In 1988 freshwater areas occupied about 1.8% or 20 million hectares (ha) of the land area of China. There are estimated to be 38,600 reservoirs (covering 800,000 ha), 20 million ponds, 2800 lakes (covering 7.3 million ha), and 6.7 million ha of river areas. Most of the aquatic areas of China are located in moderate temperate or subtropical zones. Carp thrive in the southern area while introduced trout and salmon do well in the northeast. There are extensive areas of estuaries in China that contain yellow croaker and mullet in abundance, while major rivers like the Yangtze

have sturgeon and carps. Habitat destruction and pollution of rivers are having significant impacts on river species.

China has a long history of fish farming, but the advent of intensive farming began in the late 1940s. In the 1970s, particularly with the establishment of the Chinese Academy of Fishery Sciences within the Ministry of Agriculture, the development of aquaculture and fisheries became science-based. By the 1980s, 4 million ha were producing almost 4 million metric tons of fishery products. In the late 1970s fish production was placed into "private hands" and thanks to an extensive network of 30,000 extension specialists in aquaculture and fisheries, the number of fish farmers increased to 10 million, and the production by the early 1990s exceeded 20 million metric tons. The goal for the next five-year period (1996-2000) is to increase annual production to more than 30 million metric tons.

Recent figures (1993) show that the national per capita consumption of aquatic products reached 15.5 kg, and the per capita net income of fishermen was 2,294 yuan (6 yuan/$1.00). Freshwater fish farming is concentrated around urban areas and in the southern areas such as in the flood plains of the middle to lower reaches of the Pearl and Yangtze Rivers. Genetically improved fishes, integrated agro-aquaculture techniques, improved feeds, and new disease treatments continue to enhance both the total areas for and the quantity of fish production per unit area. Net pen culture techniques and industrialized fish farming systems have been established and pearl culture techniques applied in riverine systems.

The Team was told that the culture of precious, special and high quality new species is progressing from small scale demonstration into large area popularization, and many species are being propagated at a production scale. There is occurring an "abalone culture craze", "eel culture craze" and several other kinds of "culture craze" such as the culture of mitten crab, soft-shelled turtle, and bull-frog. In 1993, 8,000 ha produced 80,000 metric tons of eel valued at 5-6 billion yuan. Bull-frog, mitten crab, and soft-shelled turtle amounted to over 30,000 metric tons. The main traditional mariculture species such as oyster, razor clam, kelp, and laver have been replaced by high-value species such as mud crab, scallop, red sea bream, grouper, and roncador in some coastal provinces.

Structure of Agricultural Biotechnology Research

In 1986, China launched a major effort to develop biotechnology research to enhance its economic development and solve major problems facing its agricultural and industrial sectors. This biotechnology initiative has three main priorities: agricultural biotechnology, medicine, and protein engineering. Given the importance of agriculture to the Chinese economy, and need to feed an expanding population, agricultural biotechnology research receives a relatively large share of China's overall biotechnology research budget.

Agricultural biotechnology research in China is well organized, and is being developed to reach a clear set of national objectives. National coordination is provided by the China National Center for Biotechnology Development (CNCBD), which is administered by the State Science and Technology Commission (SSTC). The CNCBD funds about 200 projects annually. Many of these are cross-cutting initiatives which exceed the mandate of a single Ministry. Marine and aquaculture biotechnology typically receive funding for 5-10 projects.

Agriculture biotechnology research is primarily carried out through the efforts of two ministries. The Ministry of Agriculture funds research through the Chinese Academy of Agricultural Sciences (CAAS), the Chinese Academy of Fishery Sciences (CAFS), and agricultural universities, and colleges. Aquaculture and fisheries research is funded primarily through CAFS and its 21 institutes (these institutes are described in more detail in Appendix B). There are sixteen universities receiving funding from the Ministry of Agriculture for biotechnology research, including eight "Key Universities". Shanghai Fisheries University is one of these Key Universities and was visited by the Team (see Appendix B). The State Educational Commission provides the basic funds for the institutes affiliated with the Chinese Academy of Sciences (CAS or Academia Sinica) and to state and provincial general universities. In addition to the institutes and universities funded by these two ministries, the State Commission of Planning funds six National Laboratories, each with a technical specialty. The National Laboratory working on aquatic biotechnology is the Yangtze River Fisheries Research Institute in Shashi City. The Team visited this Laboratory and it is also described in Appendix B.

Observations and Assessments of Aquatic Research Programs

Aquaculture and capture fisheries are critical production components of China's food system. The production of aquatic products is ranked fifth in importance in China's agricultural output, behind production from crops, terrestial animals, forests, and food processing. The current estimates are that 10 million farmers are annually producing more than 20 million metric tons of aquatic products. Much of the growth of the industry can be attributed to the privatization that began almost two decades ago, and to the establishment of the Chinese Academy of Fishery Sciences in 1978 - the CAFS provided the solid scientific foundation for the development of the industry.

The basis for the 9th Five-Year Plan (1996-2000) goal to boost fish and other aquatic products by another 10 million metric tons (MMT) appears to be largely by increasing the area of aquaculture production. The estimation of the areas of wetlands, including lakes and reservoirs, not currently used for aquaculture, multiplied by the current average yield per hectare (kg/ha) leads to a projected increase of 10 MMT. The application of biotechnology or other technologies to enhance current yields does not appear to be emphasized in this projected increase. Moreover, the 10 MMT projected annual increase does not appear to correct for loss of wild-caught seed (i.e., naturally produced fry and gravid adults) in these wetlands. Clearly, a certain portion of

current aquaculture production depends upon wild seed. Lastly, as China becomes more "environmental conscious" of its resources, the competition to use wetlands, lakes, and reservoirs for other uses will increase. The conclusion is that China must substantially increase its investment in the research that will enhance yields in aquatic products, and this means that aquatic and marine biotechnology will continue to gain importance.

Although the Chinese Government has made a concerted effort in the past for fisheries research, few fisheries laboratories in China are currently conducting state-of-the-art molecular biology research. Although almost all of the laboratories visited by the Team were very familiar with the techniques of molecular biology, they appeared unable to carry out certain key segments of the research due to insufficient facilities, lack of modern equipment, inability to control laboratory temperature and cleanliness, and other infrastructure limitations. The lack of adequately trained staff also was apparent.

Currently, the overall trend in funding support for fisheries research is declining. The Team was unable to obtain actual funding data, in part due to the diverse sources, but also due to the fact that Directors and Professors involved in research do not really know the magnitude of State and provincial funding. One estimate was provided for current support for fisheries research from the Science and Education Division of the Ministry of Agriculture, and that figure was "several million yuan". Admittedly, this is only one source of funding, but a crucial source for fisheries educational programs. The government policy apparently is to reduce the total number of fisheries research laboratories/institutions via reallocation of funds to the better or "Key Laboratories". Rather than actively shutting down the "de-emphasized" laboratories, it appears that they are permitted to sink or swim on their own. The observation that many of the institutes visited by the Team were pursuing alternative sources of funding fits this scenario. Indeed, the culture of highly prized and/or high-value species such as giant salamander, soft-shelled turtles, bull-frogs, and eels, or the production and sales of processed fish food, or enriched soil suggests that the Directors of these institutions have no shortage of ideas in finding "free enterprise solutions" to their funding crisis. The question that must be asked from a science perspective is whether these funds from alternative sources will be used to improve the research infrastructure and fund competitive (quality) research? The Team's impression is that these funds will prolong their existence, but only a significant reinvestment by government will prevent the eventual closure of these "de-emphasized" institutions.

China has addressed the need for developing fisheries science by supporting two parallel institutional arrangements, namely, through the establishment of the Chinese Academy of Fishery Sciences with its 21 institutions, and through the Chinese Academy of Sciences. The Team visited three major CAFS laboratories (East China Sea Fisheries Research Institute, Yangtze River Fisheries Research Institute, and the Pearl River Fisheries Research Institute), and three major CAS laboratories (Institute of Developmental Biology, Institute of Hydrobiology, and the South China Sea Institute of Oceanology). In general, the CAS laboratories were far better funded, maintained, and staffed, than the CAFS laboratories. Graduate students with active research programs could be readily seen at CAS laboratories, but rarely seen at CAFS facilities.

And, as expected, the quality research on genetic engineering of fish or shellfish was essentially confined to the CAS laboratories.

Although the Ministry of Agriculture in 1988 designated certain agricultural research facilities as "Key Laboratories", it appears that the accompanying Ministry funding was confined to buildings and equipment (infrastructure) with the staff expected to seek operating funds from diverse sources within China and from international donors. In the fisheries area, the Yangtze River Fisheries Research Institute is both a Key Laboratory identified by the Ministry of Agriculture and a National Laboratory as identified by the State Commission on Planning. This Institute has special emphasis on aquatic germplasm and aquatic biotechnology. The Ministry of Agriculture also designated "Key Laboratories" at agricultural universities. In the case of fisheries sciences, there are key laboratories at Beijing Agricultural University, Nanjing Agricultural University and Shanghai Fisheries University. The Team visited the Key Laboratory of Ecology and Physiology in Aquaculture at Shanghai Fisheries University. This Laboratory was actively conducting research on algal biotechnology, interferon gene studies, and the use of PCR for population genetics studies.

China's greatest strength in the future development of biotechnological applications to agriculture will be its human resources. An impressive system of over 50 agricultural universities is generating a large cadre of young researchers, familiar with at least some of the techniques of molecular biology. The five fisheries universities and the numerous technical schools with fishery programs provide approximately 4000 students yearly to the Nation's aquatic industries and government. However, the number of graduate students in fisheries sciences compared to other areas of agriculture (e.g., plant sciences) is disproportionately small. The Team's impression is that there are very few graduate students (especially at the Ph.D. level) affiliated with fisheries universities. Rather the students pursuing graduate training in fisheries sciences are primarily affiliated with universities having strong ties to the Chinese Academy of Sciences.

International contacts are key to the development of aquatic and marine biotechnology in China. All of the outstanding Chinese research institutions have established one or more, longstanding relationships with research institutions in the United States, Europe, Australia, or New Zealand. Leading Chinese researchers have been able to continue contacts with foreign laboratories over a number of years, often traveling outside China on a regular basis. These contacts have allowed leading Chinese biotechnology researchers access to the latest scientific information as well as access to facilities for performing types of research which are difficult to carry out in China, due to the poor infrastructure. Close contacts with foreign laboratories make it difficult, when reading published scientific literature, to determine if the work described by Chinese Scientists has been done inside or outside of China. For example, Professor Zuoyan Zhu of the Institute of Hydrobiology, Chinese Academy of Sciences, Wuhan, recently published a paper on "Growth Hormone Gene and the Transgenic Fish" in BIOTECHNOLOGY IN AGRICULTURE 1993, You, C.B. et al. (eds.): P145-155, Kluwer Academic Publishers, The Netherlands. The affiliated institutions for this work were the Departments of Molecular & Cell Biology and Zoology,

University of Aberdeen, Aberdeen, Scotland, UK. Both Professor Zhu and Professor Fangzhen Sun, Director, Institute of Developmental Biology, Beijing, did extensive post doctoral research outside of China. Both developed reputations in foreign institutions and both returned to China to assume directorships of key institutions. This certainly suggests that Chinese officials recognize the importance to the future of China of outstanding scientists who have left the country to obtain further academic and research expertise and who are enticed to return by the offer of a position as Professor and Director of a Key Laboratory.

Biosafety Guidelines

In 1994, China developed formal national oversight procedures for reviewing the biosafety of genetically modified plants, terrestrial animals, aquatic organisms, and microbes. These general guidelines were drafted by an expert committee reporting to the CNCBD and received State approval. By December 1995, each Ministry was to have detailed procedures in place for field testing and commercialization of recombinant DNA-bearing organisms. During the visit, the Team met a number of senior scientists who had contributed to the development of the detailed biosafety guidelines, although the Team did not have the opportunity to actually review the not-yet-translated guidelines developed for genetically modified aquatic organisms. However, both Drs. Kapuscinski and Hallerman briefed and shared with Chinese counterparts the USDA's Agricultural Biotechnology Research Advisory Committee's (ABRAC) Performance Standards for Safely Conducting Research with Genetically Modified Fish and Shellfish. Copies of the Performance Standards were provided to each of the laboratories visited by the Team.

Recommendations

The Team's primary recommendation is that the United States increase its interactions with China in the applications of biotechnology to aquaculture and marine programs. This strategy should encompass: (1) increased consultation on biosafety and risk assessment issues in order to harmonize oversight procedures for research and commercialization, thus ensuring market access for products produced with biotechnology, (2) increased collaboration in selected scientific and technical areas which maximize the opportunity for the United States to learn from China and (3) exploring ways to assist the U.S. agricultural biotechnology industry in gaining knowledge about opportunities for joint ventures in China.

Specific Recommendations for Follow-Up Include:

a. In the areas of biosafety and risk assessment it is recommended that:

(1) biosafety guidelines for safely conducting research with genetically modified fish and shellfish be exchanged between regulatory and scientific agencies and between scientific groups.

(2) an international conference on biosafety issues and aquaculture be jointly sponsored by the Ministry/Department of Agriculture of both countries.

(3) collaborative research projects be identified and supported on the risks of environmental release, ecological impact, and commercialization of transgenic organisms. One suggested project might be to collaborate on a well-chosen technical problem (e.g., disease resistance) in the production and evaluation of a genetically modified aquatic organism, including the challenge of integrating the modified organism into an entire genetic improvement program.

b. That the appropriate agencies, institutions, and scientists be identified to participate in the collection and preservation and exchange of aquatic germplasm to include exchange of methods to maintain and propagate rare and endangered species.

c. Explore linkages in information systems. Chinese biotechnology laboratories could benefit greatly from computerized "Silver Platter" literature search capabilities and access to other scientific information systems in the United States. Such facilities and information are available from the National Agricultural Library and university libraries. Likewise, the United States could benefit greatly from access to information about Chinese research projects. Collaboration in this area would entail technical assistance to establish a current biotechnology information database in China. For example, such a collaboration might involve an exchange of Information Specialists on a 3-month detail so as to allow time to assimilate the different systems of handling and disseminating information within a country.

d. Open a dialogue between U.S. Extension Aquaculture Specialists and Chinese Extension Aquaculture Specialists on the role of Extension in advancing the adoption of genetically modified aquatic organisms into commercial production systems.

e. Circulate this report, the reports of subsequent biotechnology activities and provide oral briefings to the U.S. scientific community, including to U.S. agricultural biotechnology companies.

APPENDIX A

SURVEY AND INFORMATION EXCHANGE ON BIOTECHNOLOGY APPLICATIONS TO FRESHWATER and MARINE ANIMAL AQUACULTURE

U.S. Scientific Exchange Team #5 to China, 6-18 October 1995

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Goals and Objectives of the Visit:

1. To receive an overview of the range of aquatic biotechnology research, development, and applications in China.

2. To understand how the Chinese address the issues of ecological risk assessment and environmental safety of aquatic biotechnology.

3. To find out what are the aquaculture goals of China in the next quarter century and how aquatic biotechnology relates to these goals.

4. To obtain from fish farmers and seafood consumers of aquatic organisms the perception of using biotechnology.

5. To explore options for collaboration between China and United States researchers.

APPENDIX A
ITINERARY AND INDIVIDUALS VISITED IN CHINA, OCTOBER 6-17, 1995

DATE	*INSTITUTION & ADDRESS*	*NAME(S)*	*TITLE*	*PHONE*	*FAX*
October 6-7	**MINISTRY OF AGRICULTURE** 11, Nongzhanguan Nanli, Beijing, 100026	Liu Congmeng, Zhang Jianqiu, Wang Donghui,	Director-General Program Officer Project Officer	86-10-419-2446 86-10-419-2759 86-10-419-2438	86-10-419-2468 "same as above" "same as above"
October 9	**INSTITUTE OF DEVELOPMENTAL BIOLOGY, CHINESE ACADEMY OF SCIENCES** Beijing, 100080	Fangzhen Sun, Yan Shaoyi, Zhong Rong Mao,	Director Professor Associate Professor	86-10-255-5140 "same as above" "same as above"	86-10-255-1951 "same as above" "same as above"
October 10	**CHINA NATIONAL CENTER FOR BIOTECHNOLOGY DEVELOPMENT** P.O. Box 8118, B7, Zaojunmiao, Haidian, Beijing 100081	Jinhu Lin, Li Qing, Wang Renwu,	Professor Assistant Researcher Chief, Animal Science	86-10-225-1986 86-10-225-1979 86-10-225-4172	86-10-225-4106 "same as above" "same as above"
October 10	**CHINESE ACADEMY OF FISHERY SCIENCES** No. 150 Qing Ta Cun South Yong, Ding Road Beijing, 100039	Si Tu Jiantong, Wang Minsheng, Li Yingren, Jilong Li,	Vice President Professor Deputy Director Associate Professor	86-10-821-444-2308 86-10-821-444-2248 86-10-821-444-2293 86-10-821-444-2271	86-10-821-4685 "same as above" "same as above" "same as above"
October 12	**SHANGHAI FISHERIES UNIVERSITY** 334 Jun Gong Road Shanghai, 200090	Li Sifa, Jixiang Wang, Wei Hua, Hu Yuan Xiang,	Professor Professor Assistant Professor Staff Member	86-21-543-109-0333 86-21-543-109-0378 86-21-543-109-0216 86-21-543-109-0345	86-21-543-4287 "same as above" "same as above" "same as above"

October 12	**EAST CHINA SEA FISHERIES RESEARCH INSTITUTE, ACADEMY OF FISHERY SCIENCES** 300 Jungong Road Shanghai, 200090	Qiao Qing-Lin, Yu Lian Fu, Gui Cong Shi, Xie Ying Liang, Gu Xin Gen,	Deputy Director & Professor Associate Professor Engineer Associate Professor Professor	86-21-543-4690 "same as above" "same as above" "same as above" "same as above"	86-21-543-4690 "same as above" "same as above" "same as above" "same as above"
October 14	**YANGTZE RIVER FISHERIES RESEARCH INSTITUTE, ACADEMY OF FISHERY SCIENCES** North Jianghan Road, Shashi, Hubei, 4340000	Laining Yu, Xingzhong Zhang, Dachun Lu, Wei Yu Sheng,	Deputy Director Professor Associate Professor Senior Engineer & Provincial Specialist	86-71-621-2541 "same as above" "same as above" 86-27-782-3882	86-71-622-8212 "same as above" "same as above" 86-27-782-3370
October 16	**INSTITUTE OF HYDROBIOLOGY, CHINESE ACADEMY OF SCIENCES** Institute of Hydrobiology Wuhan, 430072	Zhu Zuoyan, Wu Qingjinag, Liu Xiang Yu,	Deputy Director and Professor Professor Researcher	86-27-788-3481 "same as above" "same as above"	86-27-787-5132 "same as above" "same as above"
October 17	**PEARL RIVER FISHERIES RESEARCH INSTITUTE, ACADEMY OF FISHERY SCIENCES** Baihedong, Guangzhou, 510380	Liu Fiazhao, Z. Dongbiao, Wu Ruiguan, Bai Junjie,	Director Engineer Associate Researcher Associate Professor	86-88-91-543 "same as above" 86-88-91-592 "same as above"	86-88-91-531 "same as above"
October 17	**SOUTH CHINA SEA INSTITUTE OF OCEANOLOGY, CHINESE ACADEMY OF SCIENCES** 164 West Xingang Road, Guangzhou 510301	Huang Yuting, Qi-Zeng Jin, Xiang Wenzhou, Jiang Weiguo, Zhang Zhirong,	Assitant Director Professor Associate Professor Professor Professor	86-20-445-1335 "same as above" "same as above" "same as above" "same as above"	86-20-445-1672 "same as above" "same as above" "same as above" "same as above"

APPENDIX B
DETAILED REPORTS OF INSTITUTIONAL VISITS

INSTITUTE OF DEVELOPMENTAL BIOLOGY, CHINESE ACADEMY OF SCIENCES, BEIJING

The Institute of Developmental Biology was established in 1980 and is supported by the Chinese Government and the Rockefeller Foundation (with $200,000 over the next three years from Rockefeller). The Institute conducts research in both animal and plant biotechnology. The Team's visit to the Institute focused on the animal biotechnology component, with a discussion and tour of facilities led by Dr. Fangzlnen Sun, Director of the Institute. The Institute has a staff of 150, plus about 30 graduate students and post-docs, most in plant science. Although originally emphasizing basic research, a shift was made to applied research in 1988. Under Dr. Sun's leadership (since spring 1995), emphasis has been redirected to basic research in mammalian, fish, and plant developmental biology and biotechnology.

Dr. Sun addressed several aspects of developmental genetics in mammalian systems, including:

- Use of embryo splitting for purposes of cloning farm animals,
- Development and use of embryonic stem cells, with homologous recombination as a means of gene transfer, with the objective of directing secretions of pharmaceutical proteins in the milk of transgenic farm animals,
- Investigation of sperm-egg interaction, including chemical attractants and regulation of $[Ca^{++}]$ oscillation, with the possible objective of developing a novel contraceptive approach, and
- Identification of genes involved in determining cell totipotency, pluripotency, and differentiation.

Once a major research focus, research on fish developmental genetics now involves only three investigators and one graduate student, Mr. Li Shuhong. Dr. Yan Shaoyi and Dr. Zhong Rong conducts research on nuclear transfer in fish eggs, aimed at understanding interactions of egg nucleus and cytoplasm in fish development. Many combinations of donor nucleus and enucleated egg cytoplasm were examined, with two giving rise to particularly noteworthy results:

- The nucleus of common carp was transferred into enucleated crucian carp eggs, yielding a fertile generation which is now widely produced and marketed, although quantitative analysis of performance has not been performed,

- The nucleus of grass carp was transferred into the enucleated cytoplasm of blunt-nosed bream eggs, yielding 24 hybrid individuals resembling grass carp, but exhibiting morphological and biochemical differences not well characterized. Of the 24, most died, some were transferred and studied in unsecured outdoor ponds, and two individuals remain alive at the Institute.

- Triploid goldfish and mouse-rodent hybrids also have been produced.

Analysis of the hybrid fishes described has focused exclusively on morphological, chromosomal, and performance characters, and has not shifted to the molecular level. Applications of molecular genetic techniques will be necessary for progress beyond the current stage of understanding. The Team suggested the need for collaboration with other groups investigating developmental genetics of fish, for example:

- In producing zebrafish hybrids for analysis by groups investigating the molecular genetics of development, or
- Use of zebrafish-derived probes for analysis of hybrids already produced at this Institute.

Dr. Zhong briefly discussed the construction of a goldfish genomic library and ongoing efforts to clone and characterize homeobox genes using a mouse probe. It was apparent to the Team that success in the effort would depend on collaboration with Dr. Sun's group. Zhong expressed interest in transgenic fish, although his research goals were not made clear. Microinjection equipment was present in the laboratory.

The three fish development investigations conducted in this Laboratory are supported by fish holding facilities. An outdoor facility in front of the Institute comprised perhaps 30 small concrete ponds and perhaps 60 tanks holding a wide variety of fancy (exotic) goldfish lines. An aquarium facility inside the building held zebrafish and Xenopus, but was under-utilized.

Dr. Sun is an energetic, mid-career scientist who assumed the Directorship within the last year. A great effort is being devoted to much-needed renovation of laboratory infrastructure and purchase of new equipment. Dr. Sun is anxious to attract foreign-trained Chinese instructors to return to this and other Chinese institutions as senior investigators and to enhance interactions with foreign scientists. He envisions allocating some of his Institution's space to permanent laboratories for Institute scientists and some open laboratories for 3-6 year programs, involving international collaborative research and joint publications and patents. He envisions the Institute hosting foreign scientists for periods of one-to-several months, and sending staff and selected students for periodic, short-term training in foreign laboratories. He hopes for commercial applications of research on animals as bioreactors through collaboration with biotechnology firms to be housed in an institutional building to be controlled on the Institute grounds. Although Chinese government investment in facility renovation is underway, realization of the Director's vision for this Institute will depend on attracting considerable capital from outside of China.

CHINA NATIONAL CENTER FOR BIOTECHNOLOGY DEVELOPMENT, BEIJING

On October 10, 1995, the Team visited the China National Center for Biotechnology Development (CNCBD). The Center is a component of the State Science and Technology Commission. Our host for the visit was Dr. Jinhu Lin, Director, Department of Science Research and Professor; Dr. Wang

Renwu, Chief, Division for Laboratory Animal Science and Technology; and Mr. Li Qing (translator), Assistant Researcher, Director of Program 863 Preview. Dr. Ding Young, Director of CNCBD was on international travel at the time of our visit.

CNCBD is the Federal Program within the State Science and Technology Commission charged with managing and coordinating biotechnology research within China and across institutional boundaries for the purpose of economic development and social progress. CNCBD is one of five major units of a major program, the China 863 Hi-Tech R& D Program, which began in 1988 at Deng Xiaoping's direction. The five major units of the 863 Program are: Biotechnology, Information Technology (and Telecommunications), Automation Technology, Advanced Materials, and Energy Technology. It has four goals:

1). To aim for the peak of technology development and to narrow the gap between China and the developed world in high technology,

2). To train highly creative scientists and engineers,

3). To commercialize research to transform traditional industries and to develop new industries to serve development of state economy and security into the 21st century, and

4). To promote high tech development, create fertile conditions for high tech industries after the year 2000, and to prepare for steady and sustainable development.

Each technology area has an expert advisory committee. The 863 Program recognizes the importance of international cooperation, and lists 14 countries (in addition to Hong Kong) in Asia, Europe, and North America (Canada and U.S.) as cooperators. Its mandate is to develop practical policies and measures that will open ways for structuring, organizing, and managing China's science and technology. It is to do this through:

1). Unified leadership and concentration of resources [this involves appropriation of funds and assignment of jobs on the basis of proposals for specific purposes. Major issues are decided by the Hi-tech Planning, Coordinating, and Guiding Group of the State Council];

2). Effective coordination through flexible research organizations and various forms of joint R&D centers;

3). Decision making and management based on consultation with experts and specialists;

4). Training of a younger generation of scientists and technicians; discovering and improving outstanding young and middle-aged specialists;

5). Focus on developing targeted products; and

6). Initial cooperation and collaboration.

Eight Research and Development Centers have been set up, and three are Biotechnology Centers: They include the Genetic Engineering Vaccination Center (Changchun), the Genetic Engineering Drug Center (Shanghai), and the Genetic Engineering Bio-products Center (Shanghai). The goals in biotechnology are to make use of modern bioscience, with genetic engineering at the core, to (a) study and develop superior, high-yield crops and disease-resistant plants and animals, (b) conduct research in recombinant drugs and vaccines, (c) develop gene therapy for diseases, (d) carry on protein engineering and a plant genome program, and (e) use research results in developing modern biotechnology industries.

Biotechnology research on freshwater fishes and marine plants and animals began in 1985, and is quite limited; however, the new 5-year plan for 1996-2000 calls for increasing effort in this field. The investment in this field will be significant. The work will focus on developing new strains of marine and freshwater fishes, algae, crustaceans, and molluses. Control of diseases among these organisms is to be addressed.

Promoting regulation of genetic engineering so that public health, the environment, and ecological balance are protected is a responsibility of the CNCBD, which also provides overall coordination. In late 1993, it issued Safety Administration Regulations on Genetic Engineering, that will serve as a frame-work for detailed regulations that will be developed by other entities such as the Ministry of Agriculture, Ministry of Hygiene, Bureau of Oceans, Bureau of Medicine, and Bureau of Light Industry. The regulatory framework covers field experiments, laboratory research, pilot tests, industrial production, commercial release of genetically engineered organisms, and use of genetically engineered products.

As noted, CNCBD will direct policy development and interagency coordination (e.g., between Marine Bureau, Academia Senica, Bureau of Medicine, Bureau of Light Industry, and Bureau of Environment). However, each Ministry or Bureau will develop its own implementation rules. For example, the Ministry of Agriculture is developing detailed regulations regarding transgenic fish that will be published at the end of 1995. The development of marine environmental safety regulations has not yet been initiated for genetically modified marine organisms. At first, researchers strongly resisted being subjected to regulations, but their attitudes in this regard are ameliorating. The Ministry of Agriculture arranged a workshop to discuss detailed regulations. The results will be published at the end of 1995. China has limited experience in identifying potential safety problems and issues. CNCBD suggested that collaboration with the United States on these topics would result in faster progress in both countries.

In regard to research aquatic in biotechnology, efforts will involve growth enhancement, disease resistance and reproductive control, (through polyploidy and control of sex determination). The primary centers for marine biotechnology are: Qingdao Marine University; The Qingdao Marine Science Institute; Chinese Academy of Sciences; The Nanhai Marine Research Institute; Guangdong Province Freshwater Fishes Genetic Engineering Institute; Institute for Hydrobiology, Chinese

Academy of Sciences at Wuhan; and, the Heilongjiang River Freshwater Fishery Institute, Harbin, Heilongyjiang Province.

Aquaculture in China is well developed. It includes large-scale cultivation of marine algae, which are used for edible products and for production of industrial polysaccharides and pharmaceutical materials, traditional and new. In East China, production of marine plants and animals is economically important. Although few marine finfishes are cultivated, developing this field of aquaculture is important and will be supported through expanded research programs. For example, there is potential for expanded export trade in eel, but cultivation technology needs to be developed or improved. Eel diseases are a particular problem that will be addressed. Eel cultivation is a research area where China would like to collaborate with U.S., particularly in regard to reproduction and growth.

Lobster and shrimp diseases are responsible for enormous commercial losses and little is known about them. Corresponding research should focus on determining causes and isolating viruses, prevention and control of disease by developing feeds that enhance immune response, and production of disease-free stocks. For example, they would like to be able to introduce a strain with a gene for an antibacterial peptide, and they would like to cooperate with the U.S. on research to solve disease problems in shrimp. Last year, diseases in shrimp caused a loss of 4 billion yuan in one district.

Funding for research on marine species comes from four sources: 1). 863 Program (for High-Technology research) 2). National Important Research Subjects Program (important to economic development, hi-tech or not) 3). "Climbing" Project Program for high priority basic research issues, and 4). China Natural Sciences Foundation, which sponsors basic research.

The European Commission provides funds for collaborative research in biotechnology. The CNCBD houses the Chinese branch of the International Center for Gene Engineering Biotechnology. CNCBD cooperates with U.S. through the Rockefeller Foundation in regard to transgenic rice and related genome research. CNCBD manages and coordinates more than 200 biotechnology subjects on which almost 6000 Ph.D.s work. Eight or nine subjects are aquatic or marine. Marine research has been identified as one of CNCBD's six areas of focus. The six will include:

1). New varieties of domestic animals and plants with high yield, superior quality and stressing tolerance, and disease resistance,
2). New medicines, vaccines and gene therapy,
3). Protein Engineering (molecular modeling and drug design),
4). Rice Genome Program,
5). Pilot Research and Development (including new materials), and
6). Marine Research and Bioactive Marine Compounds.

Industrial development in China is primarily a government function, but the government wants research institutes to connect with companies for commercial development and cooperative efforts.

China has joint research efforts with the European Commission (EC) through the CNCBC. About five to seven projects are in progress at any one time. Research Fellows are exchanged. The EC provides funding and China sets priorities.

CHINESE ACADAMY OF FISHERY SCIENCES, BEIJING

On Tuesday, October 10, the Team visited the Chinese Acadamy of Fishery Sciences (CAFS) located in the southern suburbs of Beijing. The host for the visit was the Vice-President and Senior Engineer Si Tu Jiantong. Professor Wang Minsheng, Director of the Fishery Scientific Information Research Institute was also present. Translation was provided by Associate Professor Jilong Li (worked two years through 1988 with NOAA at Oxford, MS, and Assistant Professor Li Yingren, Division of International Cooperation.

The Chinese Academy of Fishery Sciences was founded in 1978. The Headquarters Facility in Beijing was built in 1989 and houses, in the immediate complexes, approximately 220 staff. The CAFS is funded by and reports to the Ministry of Agriculture. There are 21 Institutes within CAFS, employing 1,600 scientists and technicians, among which are 400 professors. Vice-President Si Tu described the three marine fisheries institutes, one each on the coasts of the Yellow Sea, East China Sea and the South China Sea; and the three integrated freshwater fishery institutes along the valleys of the Yangtze River, Pearl River and Heilongjiang River. In addition to these institutions, the CAFS has a Fishery Machinery and Instrumentation Research Institute and a Fishery Engineering Research Institute. The headquarters facility houses the Fishery Planning and Design Institute, the Fishery Scientific Information Research Institute and the Fishery Economics Research Institute. As previously noted, salary support for the Academy comes from the Ministry of Agiculture; however, much of the project funding comes from State (national) and Provincial sources. Members of the Academy have carried out many cooperative projects with U.S. counterparts, such as the China-U.S. Marine and Fishery Research Project.

The professional staff of CAFS have published over 3000 scientific papers in more than 400 journals. Only about 20 of the journals, however, are recognized internationally. Professor Wang Minsheng noted that the Fishery Scientific Information Research Institute has been collecting international articles and publications since 1963, and they currently receive the Fisheries and Aquatic Sciences Abstracts.

Although the Team asked about the role of biotechnology in China aquaculture and marine fisheries programs goals, the CAFS staff had little current knowledge on the progress of employing genetic engneering. Vice-President Si Tu told us that in 1984 there were 3 million fishers aquaculture aquaculturists in China. In 1994, there were 10 million, and of that number, one-third were said to be women. Only 400,000 were employed in government collective projects, suggesting that the remaining 9.6 million were privately employed. Our hosts mentioned that fishing and aquaculture are attractive lines of work because of the high market value of the products.

Responding to a question on extension, Professor Minsheng said that there were 30,000 technical extension specialists, who played major roles in rapidly transferring technology to the fisheries community. At the Province level, researchers from CAFS Institutes often transfer information directly to fish farmers via the conduct of their research on production/demonstration farms. Approximately 30 species of finfish and shellfish are cultured, but only 10 are stocked to enhance wild populations. In recent years, the focus of CAFS research on capture fisheries has shifted from natural history and productivity studies to protection and rebuilding of wild populations impacted by growing environmental problems. CAFS institutes provide data and other scientific input for fisheries management. The Ministry of Agriculture is the lead fisheries management agency. From conversations here and at the East China Sea Fisheries Research Institute, we learned that fishing is presently closed in all coastal waters of China.

Responding to a question on training, Vice President Si Tu said that there were five fisheries universities (four-year universities) and numerous technical institutes (two-year programs associated with the provinces in China). He said that fisheries science had been a defined discipline area for 35-40 years, and that currently between 3,000 and 4,000 students complete training each year. As a follow-up recommendation, we discussed the importance of exchaning information and suggested that an Information Specialist from the National Agriculture Library's Aquaculture Center and CAFS's Information Program should collaborate on information technology.

SHANGHAI FISHERIES UNIVERSITY, SHANGHAI

The Team visited Shanghai Fisheries University on October 12, 1995. We were officially welcomed by Professor Jixiang Wang of the University's International Exchange Division (IED), who briefed us on the University. Present were graduate students, He Peimin, and Kuang Mei, and Dr. Chu, Professor Li Sifa, Assistant Professor Wei Hua (Deputy Director of the Fisheries College), and Hu Yun Xiang (Deputy Director of the Key Laboratory of Ecology and Physiology in Aquaculture).

The University is under the Ministry of Agriculture. Dating from 1912, it is the oldest Chinese institution for fisheries sciences education, and has graduated 12,000 students. Its fisheries curriculum is comprehensive, offering 11 specializations at the Bachelor of Science level (e.g., ichthyology, freshwater aquaculture, fisheries products processing, and fisheries economics and management), four at the Master's level, a Ph.D. program (jointly offered with the Ocean University of Qingtao), and six two-year short-cycle courses. The University is organized into five Colleges (Fisheries Science, Engineering, Food Science, International Commerce and Trade, and Adult Education), four Departments, a 500,000 volume library and an information center. Currently, there are over 3000 students enrolled, mostly undergraduates, 41 Master's candidates, and 2 Ph.D. candidates. There are 290 faculty, including 129 professors and associate professors, and 11 research fellows. The University recently got approval for enrollment of foreign students. The University has two field stations, one for freshwater aquaculture (some 50 km from the main campus) and one for marine aquaculture (in Zhenjiang Province, 200 km away); additionally, the University has access to

cooperating government farms near the University. The University's educational budget comes from the Ministry of Agriculture, and is based on the number of students enrolled.

The Key Laboratory of Ecology and Physiology in Aquaculture was established in the 1980s and certified as an "Open Laboratory" (approved by the Governemnt to accept researchers from outside China) by the Ministry of Agriculture in 1993. The Laboratory has two professors, two associate professors, and nine junior faculty. The Laboratory is housed in a relatively new building equipped with over $1 million in funding from the World Bank, the Canadian International Development Research Center (IDRC), and the Swedish International Foundation for Science (IFS). Directed by Professor Li Sifa, a comprehensive biotechnology research program is being conducted by the Key Laboratory.

Between 1978 and 1993, Shanghai Fisheries University conducted 186 research projects that were sponsored by China's Ministry of Agriculture, the City of Shanghai, or through grants from international agencies such as the International Center for Living Aquatic Resources and Management (ICLARM, headquarters in Manila, the Philippines), IDRC-Canada, or the IFS-Sweden. Some research support has come from aquaculture enterprises, such as an Eel Culture Association from Guangdong Province. The University has established inter-institutional relationships with over 10 universities in the United States, Japan, The Republic of Korea, Canada, and the United Kingdom.

A Department of Aquaculture Technology and Biotechnology was organized this year with Dr. Chu and Dr. Wei given responsibility for program development. The first cohort of 30 students entered the program in fall 1995.

Ongoing areas of biotechnology research at Shanghai Fisheries University include the following:

- marine algae biotechnology research in Professor Jixiang Wang's Laboratory was explained by Ph.D. candidate Kuang Mei. Porphyra yezoensis is being subjected to protoplast isolation and regeneration of whole plants. Experiments are underway to introduce a bacterial marker gene. Similar lines of research have been initiated with Gracilaria asiatcia and Sarjassum sp..

- Dr. Chu described PCR work aimed at the cloning and characterization of the beta-interferon gene of fish. Understanding of cytokine action hopefully will lead to measures decreasing disease-related losses in production aquaculture.

- Professor Sifa described the major research directions of the Key Laboratory of Ecology and Physiology in Aquaculture, including:
    - Aquaculture ecosystem studies (e.g., nutrient and energy flows in ponds), in cooperation with four Japanese universities,
    - Aquatic Genetic Resources Consortia concerned with Chinese carps, tilapia, and more recently, high value species of soft shell turtle and river crab. This Consortia is supported by national, international, and corperate grants. Another important focus of this research area has been to delineate and protect the genetic biodiversity of fish species in the Yangtze River, including development of a live gene bank in oxbow lakes. The research

work has been moving into PCR-mediated analysis of mitochondrial and nuclear DNA. With funding recently lapsed for the Yangtze River fish genetics component, this line of research may prove attractive for funding by USDA's International Cooperation and Development grants program.

- Evaluation and genetic improvement of six species of tilapia, and the genetic improvement of Wuchang fish. Now in the F_4 generation, the tilapia exhibit a 15% improvement in growth rate. The breeding goal is to achieve a 20% improvement, which could be achieved within two more generations, although funding for this work has lapsed.

- The Key Laboratory publishes a biannual journal of its research papers. In 1994 and 1995, Professor Li Sifa published two major books on scientific and production aspects of freshwater aquaculture in China and is working on a new book on genetics of fishes of China.

- It was noted that the University receives no biotechnology research support through the Biotechnology 863 Hi-Tech Research and Development Program. Several potentially important lines of biotechnology research may have to be terminated for lack of funding.

Biotechnology Policy: University faculty were unaware of the government's Safety Administration Regulation of Genetic Engineering adopted by the State Science and Technology Commission in late 1993. The USDA Team explained and handed out a copy of the ABRAC Performance Standards for Safely Conducting Research with Genetically Modified Fish and Shellfish.

EAST CHINA SEA FISHERIES RESEARCH INSTITUTE, CHINESE ACADEMY OF FISHERY SCIENCES, SHANGHAI

The East China Sea Fisheries Research Insitutute (ECSFRI) was founded in 1958. It is one of the three marine institutes under the auspices of the Chinese Acadamy of Fishery Sciences (Ministry of Agriculture), and has responsibility for conduct of the basic and applied fishery sciences for the East China Sea region. Professor (and Deputy Director) Qiao Qing-Lin briefed the team on the seven research divisions of the institute:

(1) The Marine Fishery Resources Division conducts research on the biology and assessment of fishery resources, and forecasts fishing conditions, development, and use of marine resources;

(2) The Fishery Environment Research Division engages in research into hydrology and biology involved in monitoring pollution of marine environment;

(3) The Marine Fishery Research Division studies fishing methods, detection systems, and the application of new materials to fishing equipment;

(4) The Fish Processing Research Division focuses its research on preservation, processing, analysis, and comprehensive utilization of fish and shrimp;

(5) The Mariculture Research Division conducts research on the culture of marine fish and shrimp, techniques of artifical propagation and seed production, study of fish bait, and techniques to increase production;

(6) The Ichthyology Research Division is devoted to studying the taxonomy, morphology and geographical distribution of fishes;

(7) The Fisheries Scientific Information Research Division manages books and reference materials and tracks fisheries at home and abroad.

The Institute operates a fish farming station about 70 km away, where applied research on cultivation of fish and shrimp is conducted. Though the work is not carried out with commercial farmers, the results of this research are supplied to farmers. Some research oganisms are directly marketed by the fish farming station, providing a source of revenue.

Engineer Gui Cong Shi, Head of the Office of International Cooperation for the Institute (and the translator for the session) indicated that in the last decade, the Institute has gone from 460 staff (1986) to around 350 today (1995). The Institute originally had three fishery research vessels (including a 1,000 ton ocean-going ship), but recently the two smaller ships (250-ton) were sold to regional fishing companies, and the larger ship was engaged in a commercial fishing assignment off the coast of Africa.

Associate Professor Xie Ying Liang, Director of the Fisheries Information Division, noted that the Institute has published more than 1500 scientific papers and survey reports, with the large majority being published in its own journal. He shared copies from the last two years with us. He briefly described the extension activities of the Institute, indicating that 80% of the staff were involved with providing information and assistance to the regional fisheries community. Training programs for national and foreign aquaculturists occurs at the Institute.

Associate Professor Yu Lian Fu, Vice Chief of the Fisheries Resources Division, noted that the average production of shrimp through intensive cultivation programs in the brackish water of the East China Sea exceeded 4.5 metric tons/ha. The salinity of the Shanghai tidal areas is approximately 10 parts per thousand, and is ideal for many species of aquaculture interest, including: Chinese shrimp (Pennaevs chinensis), tiger shrimp (P. monodon), prawn (P. mussambers), freshwater crab, marine crab, and icefish. Hence, artifical propagation, culture technology, and breeding of shrimp constitutes a major food resource. A decade ago, the FAO sponsored the development of facilities for the investigation of disease of important species. Discussions between the Institute staff and the Team focused on the need for virus and bacterial resistance research in commercially important species. Biotechnology may offer important breakthroughs in this area, although this Institute appears not to be actively involved in this area of research. Professor Gu Xin Gen, an expert on shrimp mariculture, briefly noted that the intensive cultivation makes the need for disease control a priority research area, and that reduced stocking densities implemented this year seemed to help. Professor Qiao Qing-Lin is in charge of a new national research program on depuration of shellfish and inquired about U.S.

approaches. This is a potential area for information and expertise exchange between China and the U.S. Another area for future exchange is strategies for control of water quality of aquaculture effluents. Our hosts mentioned that the Institute has begun research on aquaculture effluents, and they asked about U.S. standards for aqauculture effluents.

Professor Qing-Lin concluded our visit by commenting on the close ties with Shanghai Fisheries University, including appointments of Institute staff to adjunct faculty, joint research activities, and research graduate (M.S. level) training. The Insitute collaborates with the Biochemistry Institute of Shanghai for shrimp disease resistance research suported through a Memorandum of Agreement with the Ministry of Agriculture. We toured the Institute's specimen collection of marine and freshwater organisms. We were told that the collection consisted of approximately 300 freshwater and 400 marine species.

YANGTZE RIVER FISHERIES RESEARCH INSTITUTE, CHINESE ACADEMY OF FISHERY SCIENCES, SHASHI CITY

On October 14, 1995, the Team traveled to the city of Shashi in the Province of Hubei and visited the Yangtze River Fisheries Research Institute. Director Yu Laining welcomed the Team and explained the structure and function of the Institute. Present were Zhang XingZhong (former Director of the Institute), Lu Dachun (Vice Director), Wei Yu Sheng, Mou Shong, Xia Hanbin, and four members of the staff not formaly introduced. Zhao Lingang of the Hubei Department of Agriculture and Animal Laboratory also was present.

Founded in 1958, the Yangtze River Fisheries Research Institute is part of the Chinese Academy of Fishery Sciences. The Institute includes 210 people, of whom 170 are working staff, 110 of whom are researchers, 35 of whom have Associate Professor or higher rank. There are four research sections: The National Key Laboratory of Freshwater Fish Germplasm Resources and Biotechnology, the Breeding and Hatchery Laboratory, the Fish Feed Laboratory, and the Environmental Resources Laboratory.

Professor Zhang XingZhong described the two main research thrusts of this key laboratory:

(1) Population genetics of Yangtze River populations of the four major Chinese carps are under investigation using enzyme and mitochondrial DNA markers, work that should be completed at this end of the 8th Five-Year Plan (1995). Quantitative genetics research addresses the characterization of strains in terms of their biology, physiology, and morphology. The purpose of this work is to promote production aquaculture and international exchange of germplasm.

(2) A second area of research emphasis is biotechnology research, involving nuclear transplantation, gynogenesis, chromosome set manipulation, gene transfer, genomic library construction, and gene cloning and characterization. Gene transfer has utilized cloned human and bovine genes,

Professor Zhang described a collaborative effort at conserving wild gene pools in situ in an oxbow lake in the Yangtze River 150 km west of Shanghai. This collaboration includes Professor Li Sifa of Shanghai Fisheries University. This Institute hosts graduate students from Shanghai Fisheries University and from the Fisheries Department of Central China Agricultural University and Hubei Agricultural College.

Funding for the Institute comes from several sources. Investigators seek competitive grant funding from the Ministry of Agriculture, as well as from the State Science and Technology Commission. Some 863 Program funding was secured in 1990, but none since, although more 863 funding is "expected" in the next fiscal-year plan. Currently, support for transgenic fish work is not included in this biotechnology funding or from the State Science and Technology Commission's other sources of funding.

The one major biotechnology research project being conducted at this Institution involved the transfer of a rainbow trout growth hormone gene (promoter not indicated) into catfish Siluris sp.. A transgenic line was established, but for lack of resources, active research was discontinued. The transgenic stock is held at the Institute's fish station. (see below)

Other biotechnology projects completed at the Institute were described in the Annual Report's for the years 1991-1994. Genomic libraries were constructed for eight species of carps (black carp, grass carp, common carp, silver carp, bighead carp, goldfish, white bream, and black bream). The growth hormone gene of black carp was cloned. A polymerase chain reaction assay was developed for a bovine growth hormone gene introduced into transgenic fish. An ELISA assay was developed for detection of fish growth hormones. While the utility of these tools for gene transfer research is clear, it was not clear that ongoing research activity utilizes them.

Research in chromosome set manipulation led to induction of tetraploidy in bighead carp and gynogenesis in common carp and grass carp. The research projects addressed nuclear transplantation and electric pulse-mediated fusion of blastula cells and fertilized or unfertilized eggs among major carps. Cryo preservation methods were developed for the sperm of black carp, grass carp, bighead carp, mirror common carp, red common carp, silver crucian carp, silver carp, and Wuchang fish. In-situ conservation as living gene pools of Wuchang fish is approached through protection of an oxbow lake of the Yangtze River. With this exception of the last mentioned item, it was unclear to the Team the degree to which these project were being pursued at present.

The Team was taken on a tour of selected laboratories of the Institution. Among the items shown were a collection of freshwater fishes including type specimens of certain species and, micro-manipulation and microinjection equipment. They also showed us an electron microscope and a variety of molecular genetics and biochemistry laboratory equipment. The Team inferred that: (1) a limited but adequate amount of fish holding facilities were present at the Institute's main building, (2) a large investment in equipment had been made in the early and mid-1980s, (3) little investment has been made more recently, and (4) that little research work was actively ongoing at present.

Biotechnology Policy

Zhang Xing Zhong, and to a lesser degree other staff, were well aware of the development of Chinese biotechnology biosafety guidelines. Zhang reported that the Ministry of Agriculture was now revising guidelines specific for fish, and that he had just returned from a meeting in Beijing concerning them. These guidelines have been developed by an aquaculture subcommittee of a science committee established by the Minister of Agriculture. The contributions of the various subcommittees following three rounds of reviews were analyzed, and will be published by the Ministry of Agriculture.

The Team presented the USDA Performance Standards for Safely Conducting Research with Genetically Modified finfish and shellfish, to which Zhang and other staff seemed receptive. The draft Chinese guidelines and the USDA Performance Standards embody similar principles. For example: (1) they focus on ecological issues, though the performance standards address genetic issues to a greater degree, (2) both agree that ecological risks poised by transgenic aquatic organisms can be controlled when experiments are conducted on a small scale, (3) experiments must be conducted in confinements that preclude escape, and (4) large-scale experiments on production posed greater risk, although it was apparent that no risk assessment research in this area was ongoing in China.

Discussion shifted to food safety aspects of agricultural biotechnology. Food safety has been considered only in a general sense in China; there are no specific guidelines in the aquaculture context. There is, however, a consenus that fish hormones would be safer than mammalian ones for human consumption, and hence, it would be well to produce transgenic fish expressing all-fish-derived gene constructs.

Rounding out the discussion of biotechnology policy, Zhang said that effecting oversight requires supervision by specific branches of government, but it was not yet clear how the oversight process would work. Aquaculture biotechnology is now in the research and development stage - commercialization is a few years away, presenting the opportunity to address such issues before commercialization.

As to the role of biotechnology in the larger picture, the ninth five-year plan calls for an increase in fisheries production from 21 to 31 million metric tons per year. The Team asked the Institute staff how this might be approached. Two general ways in which the Institute could contibute to this goal: (1) development of better-performing (i.e., faster-growing) genetic lines of fish; and (2) open more rivers, reservoirs, and lakes to aquaculture production.

In subsequent discussions, it emerged that there are ten hatcheries in China under the Ministry of Agriculture that provide broodstock for local hatcheries. Each province may have as many as 30 hatcheries where the seed stocks are provided to aquaculturists. This provides ready channels for distributing genetically improved lines for production. However, at present, parents frequently are collected from the wild, and thus, many farmers produce their own seed stock or collect it from the wild.

Field Station of Yangtze River Fisheries Research Institute,

The Team visited the Institute's field station, and was escorted by Director Yu Laining and various field staff members. This station consisted of over 60 ponds of various sizes, support buildings, and housing for field station staff. Four research activities are underway at the station, and are briefly described below:

(1) Maintenance of Pure-Species Tilapia Lines - Since tilapia frequently are produced as interspecific inbrids, it is important to maintain pure-species resource lines. A greenhouse facility was dedicated to the purpose, as it permits a method to control temperature, thus avoiding loss of fish to cold temperatures. However, this did not become known until the Team asked about the use of this greenhouse, suggesting little enthusiasm for this important industry-supporting activity by Institute staff.

(2) Supplemental Feeding of Chinese Carps - One means of intensifying carp production would be supplemental feeding. Experimental diets, composed largely of soybean and fish meals are being evaluated. The Team was surprised at this activity and questioned the wisdom of this approach to intensification of a low-input, economically efficient traditional carp polyculture system.

(3) Aquaculture of Soft-Shelled Turtles - Considerable space at the station was committed to propagation of soft-shelled turtles, more than to any other pursuit. Soft-shelled turtle production is something of a craze among Chinese aquaculturists, as turtle blood and organs are believed by many Chinese to contribute to physical stamina, supporting a market for turtle products such as pills. Small, 50-day old turtles sell for 35-40 yuan (almost $4-5 each). Four-year old turtles weigh about 500-grams and sell for about 300 yuan (almost $40). Greenhouses are being built on the station with the goal of supporting year-round growth and reducing harvest age to 16 months. Additionally, the station sells seed stock and receives considerable revenue. A system of ponds and on-site nesting sheds have been developed to promote spawning of turtles. It was clear thar the Director and staff were very enthusiastic about the turtle research and production.

(4) Transgenic Catfish - The Team had to ask about the transgenic catfish, only to learn that they are held in a poorly maintained pond lacking security againist human encroachment at the back of the station. However, brick walls were being built around key ponds to prevent theft of turtles, suggesting which projects were valued more highly.

Although 20 staff live on site, much of the station was poorly maintained. Most effort and enthusiasm centered on turtle production, a market-driven research project which provided considerable income to a cash-strapped institute. The Director suggested that some portion of the proceeds would be allocated to an internal competitive grants program. The impression gained by this Team was that without government support for aquaculture biotechnology research, market forces would direct research efforts to development of production methods for more high-valued culture species.

Discussion at the Yangtze River Fishery Research Institute brought out mention of aquaculture biotechnology research at the Freshwater Fishery Research Center at Wuxi. Xia Dequan introduced the rainbow trout growth hormone gene into Wuchang fish. The transgene was stability inherited through the F_3 generation. Growth rates were increased 60-70%. The promoter driving expression of this trait was not identified to the Team.

INSTITUTE OF HYDROBIOLOGY, CHINESE ACADEMY OF SCIENCES, WUHAN

On October 16, 1995, the Team visited to the Institute of Hydrobiology. Our host was Professor Zhu Zuoyan, Deputy Director of the Institute and who is slated to become Director in the next year. We were also hosted by Professor Wu Qingjiang who, together with Professor Zhu, works on fish genetics, including various applications of biotechnology. We also met several female and male graduate students and Liu Xiangyu, a research staff member with an M. S. degree, who were actively doing molecular genetics laboratory work during our tour of the National Key Laboratory of Freshwater Ecology and Biotechnology (FEBL). The institutional role and ongoing research of this laboratory are summarized in sections below.

Among the more than 100 institutes of the Chinese Academy of Sciences nationwide, the Institute of Hydrobiology is the only one focused on freshwater biology. Founded 60 years ago when the location was in the rural outskirts of Wuhan, the Institute is now within Wuchang, one of the tri-cities of Wuhan. The Institute's research laboratories and a set of indoor and outdoor concrete tanks (water reuse, no discharge) are located at a well maintained facility near East Lake, one of the sites of the United Nations Environmental Program's (UNEP) "Man and the Biosphere". A large indoor water-recirculating wet lab is under construction adjacent to the laboratory and tank site. Additionally, the Institute has an East Lake Ecosystem Experimental Station, which is an experimental fish culture station having more than 100 ponds of various sizes, and is located 6 km away on the other side of East Lake. The Institute has an even larger experimental station in Guangdong Province.

The Institute has seven Departments (Ichthyology, Fish Genetics and Breeding, Fish Pathology, Freshwater Ecology, Phycology, Water Pollution Biology, and the River Dolphin Research Department); a National Key Laboratory for Freshwater Ecology and Biotechnology (FEBL); a freshwater biodiversity station, one of 17 such stations nationwide; and the largest museum of freshwater fishes in Asia, whose collection includes 900 species, 300 of which are from the Yangtze River, and 200 type specimens. As a National Key Laboratory established in 1989, the FEBL is a national priority for research support (e.g., the new wet lab under construction) and is open to visiting researchers from China and abroad.

The Institute has 400 staff, 170 retired staff living on-site, a number of graduate students from various universities working on thesis research, and over 50 post-graduate students. With encouragement from recent government reforms, the Institute has added a system of rotating post-graduates through the Institute, while retaining the traditional system of tenure for much of the staff.

Professors at the Institute give periodic lectures at several universities in addition to supervising the thesis research of graduate students who come to the Institute from various universities. Institute researchers also collaborate with researchers of other organizations, such as the Molecular Biology Center in Shanghai and various institutes of the Chinese Academy of Fishery Sciences, via specific research projects funded by the national government.

Basic operating funds and seventy percent of staff salaries comes from the Chinese Academy of Sciences. Over 140 ongoing research projects are supported by the Natural Science Foundation, other domestic sources (e.g., provincial and city government programs), the national 863 Program (e.g., Professor Zhu's research on transgenic fish), the State Key Laboratory Developmental Program (e.g., a shrimp disease project involving several cooperating institutions), and the 921 Program for research in outer space.

Summary of Research underway at the National Key Laboratory of Freshwater Ecology and Biotechnology: This is a dynamic, multi-disciplinary laboratory operating under the "open lab" policy, and has already hosted a number of outside post-doctorates and established experts for research visits of varying lengths. Freshwater ecology research includes work on energy transfer, nutrient cycles (material metabolism), and interrelations among various ecosystem components. Due to the brevity of our visit at the Institute, the Team did not have a chance to tour and learn more about this part of FEBL. The biotechnology section of FEBL has five research groups: gametogenesis and gamete engineering, gene engineering, crustacean genetics, somatic cell genetics, and cytogenetics and population genetics. A common goal of much of the biotechnology research is to develop improved or new varieties of fish and shellfish (e.g., growth-enhanced lines and disease resistant lines) to help boost aquaculture production in China. As an example of ongoing projects showing potential towards this goal, the Laboratory has developed a faster-growing strain of all-female common carp via gynogenesis and sex-reversal and has distributed fingerlings throughout China. Because of the brevity of our visit, the Team's tour of the research laboratories touched only on the following ongoing projects:

- Transgenic Fish - Zhu's group is an international leader in production and evaluation of physiology and performance of trangenic fish. They have F_1, F_2, and F_3 generation transgenic common carp (red carp) expressing a human growth hormone gene (hGH) driven by the mouse metallothionein-1 (MT) promoter. In fully contained indoor tanks, they have demonstrated physiological function of the hGH - transgene being expressed in tissues other than the pituitary via comparisons between hypophysectomized transgenic F_2 fish (growth continued after pituitary removal) and non-transgenic F_2 fish (growth ceased after pituitary removal) (Cui and Zhu 1993, Fish Physiology and Biochemistry 12(2):161-169); significantly higher specific growth rates (both wet and dry weight), energy and protein content, and conversion efficiencies (in terms of wet weigh, dry weight, and protein) of the F_2 transgenic than non-transgenic controls (Cui et al. Chinese Science Bulletin: in press, English language); significant differences between energy budgets of F_2 transgenics and controls, with transgenics channelling a higher preparation of energy to growth and fecal production and a lower proportion to nitrogenous waste and metabolism and significantly higher protein content of dry matter (Cui et al, Chinese Science

Bulletin, in press). After 263 days growth in ponds (at the Institute's isolated experimental pond facility in Guangdong Province), this same line of F_2 transgenics had a different frequency distribution of wet weights. compared to controls, with higher mean and a skew towards the higher tail of the distribution (unpublished data). Also, they have indications of between-family variation in stability of integrated transgenes (most fish bear 2-6 copies/cell and maximum number = 6400 copies/cell) and lower than expected transmission rates to progency from the following crosses:23% for wild-type x transgenic founders; 22% for wild-type x F_1 transgenics; and 66% of F_1 transgenics x F_1 transgenics.

This group also has developed anitbodies to a carp GH which they have used to show that a small, localized region of the grass carp anterior pituitary secretes GH. Finally, they have ongoing studies on the characterization and cloning of other fish genes (e.g. grass carp GH gene, Zhu et. al. 1992. Eur. J. Biochem. 207:643-648), and transfer of additional gene constructs into a variety of species (e.g. loach, crucian carp, and grass carp).

- Chromosome - set manipulations: Wu's group has induced triploidy in crucian carp and characterized the meiotic processes and products responsible for the natural occurrence of triploid gynogenetic common carp. Extension workers have put these fish into 666, 000 m^2 of fish pond farming in Hubei Province. They also have developed a fast-growing line of crucian carp which they propagate via gynogenesis and have distributed to fish farms throughout China. This group is also working on tetraploidy induction in goldfish towards the goal of mating them with diploids to yield sterile tripolids.

- Monosex populations: Given that common carp females grow faster than males, Wu and colleagues have develop an all - female line (red carp females x sex-reversed mirror carp males) and have distributed fingerlings to farms throughout China.

- Population genetics: Various studies to characterize population genetic structure of fish and crustacean species are underway. Analytio methods include isozyme polymorphisms, mtDNA, RFLPs, PCR analysis (primarily involving RAPDs), and DNA fingerprinting. Mr. Liu expressed interest in obtaining primers for investigating microsatellite polymorphisms.

- Somatic cell genetics: The goal of this line of research is to stop aquaculture losses due to grass carp viral disease by developing viral-resistant lines. Intending to benefit from presumptive cytoplasmic resistance factors, this group has successfully implanted somatic cell nuclei from crucian carp fin tissue into enucleated eggs of red carp, yielding clonal lines of normally developed fish.

- Crustacean genetics: Current research addresses questions in population genetics, fertilization, biology, and cell engineering. Two specific examples are cryopreservatoin of certain Artemia life-stages and induction of triploidy in Eriocheir heplinensis.

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Role in Biosafety for Aquatic Genetically Modified Organisms: None of the Institute's researchers serve on the National Advisory Committee to the Ministry of Agriculture (MOA), which is currently developing specific rules for biosafety of genetic engineering in agriculture, including a specific section for aquatic organisms. The MOA has consulted Zhu and other staff members, however, for scientific input on this issue. A goal of the biotechnology staff at FEBL is to perfect the production of sterile transgenic fish so that only sterile individuals would be released to production fish ponds in the future. This group of researchers is poised to conduct experiments for filling important knowledge gaps on biosafety of growth - enhanced trangenic fish. They have in hand two essential elements for such research: a documented faster-growing line of trangenic carp, with the added benefit that a number of ecologically important bioenergetic traits have already been characterized in the F_2 generation; and confinement facilities appropriate for propagating and rearing carp, with capabilities of simulating some natural ecological conditions. Regarding the latter, the existing concrete ponds both indoors and outdoors at the Institute's main building have no discharge to natural waters, are run as static systems with slow flow through a water-reuse system and replacement of evaporated water, and with screening of all drains. Two additional facilities with potential for confined biosafety research are the new indoor wet laboratory under construction, which will be a water-reuse system, and the 100 pond site 6 km away, which could be confined at a reasonable cost. Thus, discussions with our hosts at FEBL identified biosafety research as one among several good prospects for collaboration with U.S. scientists.

PEARL RIVER FISHERIES RESEARCH INSTITUTE, CHINESE ACADEMY OF FISHERY SCIENCES, GUANGZHOU

The Pearl River Fisheries Research Institute of the Chinese Academy of Fishery Sciences was founded in 1979, but its work began as early as 1953 under the aegis of the South China Sea Fisheries Research Institute. The Institute is commissioned as a comprehensive research institution for regional freshwater fisheries in China. The Institute covers 20 hectares and has good environmental conditions and a good water supply from the river which is naturally controlled to ponds by tidal flow.

The Institute conducts research and development programs, and education and outreach programs to aquaculture farmers. In 1958, it was the first Institution to artificially propagate Chinese carps. It also developed a vaccine for grass carp that increased survival to 80%. It has conducted international training classes in aquaculture under the sponsorship of the United Nations. Its research programs focus on all aspects of freshwater fish culture. The Institute works with about 20 species of fish. Its 200 member staff includes 100 researchers whose are organized into five divisions:

- Breeding and Culture Division,
- Fish Diseases and Control Division,
- Nutrition and Physiology Division,

- Bioengineering/Biotechnology Division, and
- Environmental Protection Division.

The Institute has been designated a "Key Laboratory" under the Ministry of Agriculture and is a branch monitoring station of the Chinese Environmental Monitoring Center. The monitoring includes water chemistry and pollution. The Institute's specimen collection includes about 300 species, mostly of the family Cyprinidae.

In the Bioengineering/Biotechnology Division, the work focuses on two primary areas: (1) development of polyploids, and, (2) the development of transgenic fishes. In the second area they have cloned fish growth hormone genes into bacteria for production and subsequent use by injection and feeding. The Institute is especially interested in producing a transgenic mud carp that is more tolerant to cold temperature. They hope to increase its tolerance by at least 2° C which would significantly expand the area in which species can be cultured. One approach in this work is microinjection of DNA from a more cold-tolerant carp into a mud carp. The researchers may be under the erroneous impression that introducing the anti-freeze gene of flounder will increase cold tolerance in temperature ranges above freezing. However, language problems did not allow this point to be adequately explored. In any case, the researchers have succeeded so far in increasing the tolerance range of the mud carp to 6° C from 7-8° C. They promised to mail us papers on this research achievement. The scientists recognized that there is much to do in order to understand mechanism of cold tolerance. The Institute maintains formal and informal ties and collaborations with other researchers through, for example, the Chinese Aquaculture Association and the Network of Aquaculture Centers in Asia. Students from educational institutions conduct research and thesis projects at the Institute. There are many institutions in Guangzhou that are appropriate for cooperation or collaboration.

The Institute has no 863 Funds, but it does have funding from the Natural Sciences Foundation. Funding for the Institute comes from two sources:

1. Government
    a. operating funds (salaries, etc.)
    b. capital improvement
    c. research funding through competition from many sources
2. Institute
    a. generated funds from sale of (formulated) feeds

Pollution is reducing populations of some species, although water quality standards for effluents from industry are enforced by a system of penalties. Losses of migratory species [e.g., Clupeids] is caused in part by construction of dams. The Director of the Institute would like to exchange fish specimens with U.S. institutions. The Team expressed interest in arranging for the exchange of specimens between the Institute and the University of Minnesota and Virginia Polytechnic Institute and State University.

Thirty percent of China's production is based on mud carp, which can be used for an exported, canned product. The Institute's concrete hatchery and breeding ponds for Chinese carps include large circular (4-meter diameter) ponds for egg laying and small, rapid flow, concentric circular ponds for hatching. The Institute uses hormone injections to induce spawning. One staff member can manage the fry production system, which can produce up to $1x10^6$ larvae in one small 2.5 m pond.

The Institute is definitely interested in cooperative work with U.S. scientists and asked for ideas. Disease detection and control would be one appropriate area.

SOUTH CHINA SEA INSTITUTE OF OCEANOLOGY, CHINESE ACADEMY OF SCIENCES GUANGZHOU

The South China Sea Institute of Oceanology is a component of Academia Sinica (the Chinese Academy of Sciences), and was established in Guangzhou in 1959. In the absence of the Director (Professor Pan Jinpai), the Team was hosted by the Assistant Director and Senior Engineer Huang Yuting. A 20 minute video was shown introducing the Institute. The South China Sea Institute is one of two Institutes of Oceanology in China. It is comprised of 10 laboratories with 700 staff, two research vessels and four experimental sites. The overall mission of the Institute is to conduct research on tropical marine environmental resources and oceanography problems in ocean resource exploration. The majority of the Institution's resources and staff are devoted to physical oceanography, marine tectonic geophysics, marine sedimentology, marine chemistry and physics, and marine instrument applications. The biological programs and projects are primarily conducted at the four experiment stations (Zhanjiang, Shantou, Hainan, and Daya Bay), and involve fundamental biological studies on the impact of pollution on marine organisms. The Team was briefed on four current biological projects occurring at the Institute. These were (1) cultivation of pearl forming scallops; (2) production of polysaccharides and phytochemicals with medicinal activity; (3) reproductive and life cycle studies on the giant salamander; and (4) the influence of ultrasound, electromagnetic fields and laser irradiation on the hatchability, embryonic development, and growth of goldfish (Carassius auratus) and loach (Paramisgurnus dabryanus).

Professor's Jiang Weiguo and Jin Qi-Zeng described their work on triploid scallops for pearl production. Owing to reduced gonadal development, it is easier to implant pearl nuclei; pearl retention rates are increased, pearl growth rates are faster, and harvested pearls are larger and show less dark discoloration than observed in diploid scallops. Pilot-scale testing of triploids is underway in collaboration with some breeding farms. Before commercializing this technology, Jiang explained that they need to increase the low rates of triploidy induction yielded by chemical induction. They already know that heat or pressure shock do not increase the percentage of triploid individuals. Thus, they are now conducting studies on induction of tetraploidy. If this succeeds, the next step would be to cross fertile tetraploids with diploids, which could yield 100% triploid offspring. Team member Eric Hallerman suggested that Jiang contact Dr. Guo Ximing of Rutgers University, who has found ways to increase production rate of tetraploids in oysters.

Associate Professor Xiang Wenzhou described his work on development of products from Spirulina and other microalgae. He has successfully adapted a freshwater form of Spirulina to seawater. The motivation for this is that Spirulina has a higher protein content when raised in seawater compared to freshwater. This product is already marketed domestically as a health food tablet called "Sea Power" (translated brand name). His laboratory is also conducting studies to improve extraction from microalgae of a blue coloring compound, phytocyanin, and to increase levels of polysaccharides with presumed anti-cancer properties. This group is interested in eventually inserting cold tolerance genes into their seawater-adapted line of Spirulina. The Team notes that such work would pose a number of biosafety issues. Confinement of transgenic Spirulina would be particularly difficult because of the small size of organisms and the open - system nature of most seawater aquaculture operations.

Professor Zhang Zhirong directs research in the Biophysical Laboratory established in 1984. The laboratory investigates effects of magnetic fields, laser beams, and ultrasound on the performance of captive-reared organisms. One aspect of her work has been to improve captive breeding of a number of species on the brink of extinction, including the giant salamander, jinggian tortoise and Chinese sturgeon. The Team toured an underground rearing facility holding captively bred giant salamanders of various ages and sizes. Professor Zhang's research is of a non-traditional approach. The biological effects of low-frequency electric and magnetic fields (ELF-EMF) is a controversial area in the United States. It is known that low level ELF-EMF exposure may influence cell proliferation, and from the development of effective therapeutic applications of pulsed electromagnetic fields, there are reports of EMF-promoted bone healing in human patients. The reproducibility and scientific acceptance of Professor Zhang's unique research remains to be determined.